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Effect of type of fiber, site of fermentation, and method of analysis on digestibility of soluble and insoluble fiber in rabbits[1]

R. Abad-Guamán, R. Carabaño, M. S. Gómez-Conde, and J. García[2]

Departamento de Producción Animal, E.T.S.I. Agrónomos,
Universidad Politécnica de Madrid, Ciudad Universitaria, 28040 Madrid, Spain

ABSTRACT: The effect of type of fiber, site of fermentation, method for quantifying insoluble and soluble dietary fiber, and their correction for intestinal mucin on fiber digestibility were examined in rabbits. Three diets differing in soluble fiber were formulated (8.5% soluble fiber, on DM basis, in the low soluble fiber [LSF] diet; 10.2% in the medium soluble fiber [MSF] diet; and 14.5% in the high soluble fiber [HSF] diet). They were obtained by replacing half of the dehydrated alfalfa in the MSF diet with a mixture of beet and apple pulp (HSF diet) or with a mix of oat hulls and soybean protein (LSF diet). Thirty rabbits with ileal T-cannulas were used to determine ileal and fecal digestibility. Cecal digestibility was determined by difference between fecal and ileal digestibility. Insoluble fiber was measured as NDF, insoluble dietary fiber (IDF), and in vitro insoluble fiber, whereas soluble fiber was calculated as the difference between total dietary fiber (TDF) and NDF (TDF–NDF), IDF (TDF–IDF), and in vitro insoluble fiber (TDF–in vitro insoluble fiber). The intestinal mucin content was used to correct the TDF and soluble fiber digestibility. Ileal and fecal concentration of mucin increased from the LSF to the HSF diet group ($P < 0.01$). Once corrected for intestinal mucin, ileal and fecal digestibility of TDF and soluble fiber increased whereas cecal digestibility decreased ($P < 0.01$). Ileal digestibility of TDF increased from the LSF to the HSF diet group (12.0 vs. 28.1%; $P < 0.01$), with no difference in the cecum (26.4%), resulting in a higher fecal digestibility from the LSF to the HSF diet group ($P < 0.01$). Ileal digestibility of insoluble fiber increased from the LSF to the HSF diet group (11.3 vs. 21.0%; $P < 0.01$), with no difference in the cecum (13.9%) and no effect of fiber method, resulting in a higher fecal digestibility for rabbits fed the HSF diet compared with the MSF and LSF diet groups ($P < 0.01$). Fecal digestibility of NDF was higher compared with IDF or in vitro insoluble fiber ($P < 0.01$). Ileal soluble fiber digestibility was higher for the HSF than for the LSF diet group (43.6 vs. 14.5%; $P < 0.01$) and fiber method did not affect it. Cecal soluble fiber digestibility decreased from the LSF to the HSF diet group (72.1 vs. 49.2%; $P < 0.05$). The lowest cecal and fecal soluble fiber digestibility was measured using TDF–NDF ($P < 0.01$). In conclusion, a correction for intestinal mucin is necessary for ileal TDF and soluble fiber digestibility whereas the selection of the fiber method has a minor relevance. The inclusion of sugar beet and apple pulp increased the amount of TDF fermented in the small intestine.

Key words: digestibility, insoluble dietary fiber, intestinal mucin, rabbit, soluble dietary fiber

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INTRODUCTION

Soluble fiber is not usually measured in rabbit diets in spite of its positive effect on rabbit health (Trocino et al., 2013). This circumstance is explained by the higher complexity of the available methodology for soluble fiber compared with that of insoluble fiber and the lack of agreement on the adequate methods, because most of the methods do not resemble the physiological conditions (Monro, 1993). Soluble dietary fiber can be

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[2]Corresponding author: javier.garcia@upm.es

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determined as the difference between total dietary fiber (**TDF**) and the insoluble fiber (Van Soest et al., 1991). The soluble fiber calculated as the difference between TDF and NDF (**TDF–NDF**) is commonly used (Xiccato et al., 2012), but this value is higher than the value obtained from other methods (Abad et al., 2013).

The benefit of soluble fiber on rabbit health might be related to the improvement of gut barrier function in the small intestine and the fermentability of this fraction along the digestive tract (Trocino et al., 2013). Therefore, the determination of the ileal and cecal digestibility of soluble fiber would be of interest. Furthermore, the nutritional meaning of each method for soluble fiber determination might be different and worthy of being clarified. Previous results in rabbits (Gidenne, 1992) and pigs (Jørgensen et al., 1996; Wilfart et al., 2007) reported poor degradability of plant cell walls before the cecum according to their low or even negative ileal digestibility. These results might be related to the presence of endogenous carbohydrates in ileal digesta that would be recovered in TDF analysis. Abad et al. (2013) reported that intestinal mucins are partially retained in the TDF residue, leading to an overestimation of ileal TDF content, and proposed a simple correction.

The aim of this work was to identify the effects of type of fiber (low, medium, and high soluble fiber level), site of fermentation, method to quantify insoluble and soluble fiber, and correction of the intestinal soluble fiber content for intestinal mucins on the digestibility of fiber fractions in rabbits.

MATERIALS AND METHODS

Diets

Three diets with different soluble fiber content (determined as the difference between TDF and NDF) were formulated (Table 1). The low soluble fiber (**LSF**), medium soluble fiber (**MSF**), and high soluble fiber (**HSF**) diets contained 8.5, 10.2, and 14.5% of soluble fiber (on DM basis), respectively. The MSF diet had dehydrated alfalfa as the main source of fiber. The other 2 diets were formulated by replacing half of the dehydrated alfalfa used in the MSF diet by a mixture of beet and apple pulp (75:25; HSF diet) or by a combination of oat hulls and soya bean protein concentrate (Soycomil P-economy; Loders Croklaan, Wormerveer, The Netherlands; 88:12; LSF diet). These 3 diets were formulated to contain 12.5 MJ DE/kg DM and 12 g digestible CP/MJ DE according to the nutrient requirements of breeding does (de Blas and Mateos, 2010). They also had similar levels of starch and CP and TDF, which ranged from 40.9 to 42.5% DM (Table 1). Additionally, they contained robenidine hydrochloride (Cycostat; Zoetis, Paris, France) and 0.5% of alfalfa meal labeled with Yb_2O_3 as indigestible marker (Udén et al., 1980).

Animals and Housing

Thirty New Zealand White × Californian rabbit does weighing 4,555 ± 63 g were surgically fitted with a glass T-cannula at the ileum, 10 to 15 cm before the ileocecocolic junction, according to the procedure described by Gidenne et al. (1988) and in compliance with the Spanish guidelines for the care and use of animals in research (Spanish Royal Decree 1201/2005; BOE, 2005). After 6 wk of recovery, rabbits reached their previous level of ADFI and were ready to begin the trial. Animals were individually housed in wire metabolism cages measuring 405 by 510 by 320 mm that allowed feces collection. Rabbits had ad libitum access to feed and water. The housing conditions were controlled during the whole experimental period as follows: a 12 h light:dark cycle was established, the light was switched on at 0730 h, and temperature was kept between 15 and 24°C by heating and cooling systems combined with continuous forced ventilation.

Experimental Procedure

Animals were randomly allotted to each experimental diet (10 rabbits/diet). After 14 d of adaptation period to the diets, the feed intake was recorded and the whole fecal excretion was collected for 4 consecutive days. Following the hard feces collection, 2 ileal samples from each rabbit were gathered under gravity for 1 h during 2 consecutive days (1/d) and mixed together. Both ileal samples were collected from 1900 to 2100 h to avoid the effect of the soft feces excretion period on the protein ileal flow (Merino and Carabaño, 2003). Fecal and ileal samples were stored at –20°C, frozen dried, and ground to 1 mm for further analysis. The samples were individually ground to determine DM, Yb, ash, GE, CP, starch, TDF, insoluble dietary fiber (**IDF**) according to the AOAC (AOAC, 2000), NDF, 2-step in vitro pepsin/pancreatin DM indigestibility (in vitro insoluble fiber), and soluble fiber calculated as the difference between TDF and IDF (**TDF–IDF**), soluble fiber calculated as the difference between TDF and in vitro insoluble fiber (**TDF–in vitro insoluble fiber**), or TDF–NDF. The apparent fecal digestibility of DM and dietary components (OM, GE, CP, starch, TDF, insoluble fiber, and soluble fiber) were determined according to the European reference method (Perez et al., 1995). The apparent ileal digestibility of DM and dietary components (OM, CP, starch, TDF, insoluble fiber, and soluble fiber) were determined by the dilution technique using Yb as a marker as follows:

Table 1. Ingredient and chemical composition of experimental diets (low soluble fiber, medium soluble fiber, or high soluble fiber diet)

Item	Type of fiber		
	Low soluble	Medium soluble	High soluble
Ingredient, % as fed basis			
Dehydrated alfalfa	13.9	28.3	13.9
Oat hulls	14.7	–	–
Beet pulp	–	2.30	15.0
Apple pulp	–	–	5.00
Soybean meal concentrate	2.00	–	–
Boiled wheat	32.3	32.3	32.3
Wheat bran	8.40	8.40	8.40
Sunflower meal	7.10	7.10	7.10
Soybean meal	11.1	11.1	11.1
Sunflower hulls	4.40	6.00	4.40
Lard	3.30	2.30	0.50
Dehydrated alfalfa mordanced with Yb	0.50	0.50	0.50
L-lysine HCL	0.45	0.40	0.45
DL-methionine	0.10	0.10	0.10
L-threonine	0.15	0.10	0.15
Sodium chloride	0.50	0.60	0.40
Calcium oxide	0.60	–	0.20
Mineral/vitamin premix[1]	0.50	0.50	0.50
Analyzed chemical composition, % DM			
DM	91.8	91.4	91.7
Ash	6.54	7.10	6.72
CP	19.9	20.3	19.7
CP-TDF[2]	5.21	5.30	4.02
CP-IDF[3]	2.40	2.21	1.74
CP-NDF[4]	3.44	4.20	3.72
CP-in vitro insoluble fiber[5]	5.12	5.20	3.84
Ether extract	5.91	5.53	3.74
Starch	21.1	20.8	20.5
TDF	41.4	40.9	43.5
Insoluble fiber			
IDF	33.6	30.9	30.4
NDF	32.9	30.7	29.0
In vitro insoluble fiber	31.8	30.9	30.2
ADFom[6]	16.2	16.4	16.9
ADLom[7]	3.92	4.71	3.74
Soluble fiber			
TDF–IDF[8]	7.82	9.96	13.1
TDF–NDF[9]	8.54	10.2	14.5
TDF–in vitro insoluble fiber[10]	9.61	10.0	13.3
GE, MJ/kg DM	19.1	19.0	18.6
Calculated chemical composition,[11] % DM			
Lysine	1.26	1.18	1.22
Methionine	0.41	0.41	0.40
Methionine + cysteine	0.72	0.72	0.70

[1]Provided by Ibérica de Nutrición Animal S.L. (Madrid, Spain). Mineral and vitamin composition (per kg of complete diet): 40 mg of Mn as MnO, 50 mg of Zn as ZnO, 1.25 mg of I as KI, 40 mg of Fe as $FeSO_4$, 25 mg of Cu as $CuSO_4$, 0.5 mg of Co as $CoSO_4$, 250 mg of choline chloride, 2 mg of riboflavin, 5 mg of calcium D-pantothenate, 15 mg of nicotinic acid, 1 mg of menadione sodium bisulfite, 9,000 IU of vitamin A as retinyl acetate, 1,800 IU of vitamin D_3 as cholecalcipherol, 12.5 IU of vitamin E as **α**-tocopherol acetate, 0.01 mg of cyanocobalamin, 1 mg of thiamine, and 66 mg of robenidine hydrochloride (Cycostat; Zoetis, Paris, France).

[2]CP-TDF = CP linked to total dietary fiber (TDF).

[3]CP-IDF = CP linked to insoluble dietary fiber (IDF).

[4]CP-NDF = CP linked to NDF; NDF = **α**-amylase neutral detergent fiber corrected for ash.

[5]CP-in vitro insoluble fiber = CP linked to in vitro insoluble fiber.

[6]ADFom = ADF corrected for ash.

[7]ADLom = ADL corrected for ash.

[8]TDF–IDF = soluble fiber calculated as difference between TDF and IDF.

[9]TDF–NDF = soluble fiber calculated as the difference between TDF and NDF.

[10]TDF–in vitro insoluble fiber = soluble fiber calculated as difference between TDF and in vitro insoluble fiber.

[11]Calculated values according to Fundacion Española para el Desarrollo de la Nutrición Animal (De Blas et al., 2003).

ileal apparent digestibility of DM = [1 – (dietary Yb concentration/ileal Yb concentration)] × 100 and

ileal apparent digestibility of dietary components = [1 – (dietary Yb concentration × ileal component concentration/ileal Yb concentration × dietary component concentration)] × 100.

Furthermore, the intestinal crude mucin in ileal and fecal digesta was determined to express TDF and soluble fiber without intestinal mucins (Abad et al., 2013). The cecal digestibility of each chemical constituent was calculated for each rabbit as the difference between the fecal and ileal digestibility. Cecal digestibility of mucins was determined for each rabbit as the difference between mucin ileal flow and mucin fecal excretion.

Analytical Methods

Procedures of the AOAC (2000) were used to determine DM (method 934.01), ash (method 942.05), CP (method 968.06), ether extract (method 920.39), starch (amyloglucosidase-α-amylase method; method 996.11), TDF (method 985.29), and IDF (method 991.42). Dietary NDF was determined using the filter bag system (Ankom Technology, Macedon, NY) according to Mertens et al. (2002), and a thermostable amylase without any sodium sulfite was added. It was corrected for its own ash and protein as indicated for IDF. Dietary ADF and ADL were analyzed according to the AOAC (2000; method 973.187) and Van Soest et al. (1991), respectively. The in vitro insoluble fiber was performed in Ankom bags (Ankom Technology), and the indigestible residue was corrected for CP and ash (Abad et al., 2013). The soluble fiber was calculated by difference as TDF–IDF, TDF–in vitro insoluble fiber, or TDF–NDF. Gross energy was determined by adiabatic calorimetry. Diets were analyzed in triplicate, and ileal digesta and feces were analyzed in duplicate. One gram of ileal content and 3 g of feces from each rabbit were used to analyze their crude mucin content, which was determined using the method of precipitation with ethanol (Leterme et al., 1998; Romero et al., 2011) and using pectinase (Sigma P2401; Sigma-Aldrich, St. Louis, MO) to remove soluble fiber (Abad et al., 2013). Crude protein on fecal mucin was determined using only 1 pooled sample for each treatment due to the small amount of residue obtained. Additionally, the Yb content of diets and ileal digesta were analyzed by atomic absorption spectrometry (Smith Hieftje 22; Thermo Jarrel Ash, Andover, MA; García et al., 1999).

Statistical Analysis

Repeated measures analysis was used to analyze the results obtained for apparent ileal and cecal digestibility using a mixed model (SAS Inst. Inc., Cary, NC). The model for DM, OM, and starch digestibility included, as fixed sources of variation, the soluble fiber level (diet), the site of digestion (ileum vs. cecum), and their interaction. The model for CP and TDF digestibility included, as fixed sources of variation, the soluble fiber level (diet), the site of digestion/fermentation, the mucin correction, and their interactions. The model for insoluble fiber digestibility included, as fixed effects, the soluble fiber level (diet), the site of fermentation, the method, and their interactions. The fixed effects of the model for soluble fiber digestibility were the soluble fiber level (diet), the site of fermentation, the method, the mucin correction, and their interactions. To compare the insoluble with the corrected soluble fiber digestibility, a model was used containing soluble fiber level (diet), site of fermentation (ileum vs. cecum), fiber fraction (insoluble vs. soluble), and method as fixed sources of variation. The model for daily fermented fiber (once corrected for mucins) included DMI as covariate and the soluble fiber level (diet), site of fermentation (ileum vs. cecum), fiber fraction (insoluble vs. soluble), and method as fixed sources of variation. In all cases, the rabbit was included as a random variable in the model. A compound symmetry structure was fitted because it showed the lowest value of the Schwarz Bayesian criterion (Littell et al., 1998).

The results obtained for fecal digestibility were analyzed using a mixed model considering the soluble fiber level (diet) as the main source of variation for DM, OM, starch, and GE digestibility. The model for fecal CP and TDF digestibility included, as fixed sources of variation, the effect of the soluble fiber level (diet) and the mucin correction; for insoluble fiber digestibility, the model included the soluble fiber level (diet) and the method; and for the soluble fiber digestibility, the model included the soluble fiber level (diet), method, and mucin correction.

The model for the ileal flow of dietary components had, as source of variation, the soluble fiber level (diet) and the DMI as a covariate and for insoluble and mucin corrected soluble fiber also contained the method. In these models, all the interactions among the fixed factors were also considered. The data are presented as least squares means, and they were compared using a protected *t* test. Linear regressions between all measurements of the amount of insoluble and soluble fermented fiber using different methods were determined including feed intake as covariate.

RESULTS

The DMI did not differ among rabbits fed different dietary treatments (151, 141, and 132 g DM/d for the LSF, MSF, and HSF diets, respectively; SEM = 10.4, $P = 0.44$; data not shown). In rabbits fed the HSM diet, the average of ileal and cecal digestibility of DM and OM increased by 11% compared with the other 2 groups ($P < 0.01$; Table 2). However, the type of diet did not influence ileal and cecal DM, OM, and starch digestibility. In the ileum, 75% of the fecal digestible DM and OM was digested and almost all of the fecal digestible starch (99%; $P < 0.01$) was digested. However, fecal DM, OM, and GE digestibility of rabbits fed the HSF diet were 9% higher than those of rabbits fed the other 2 diets ($P < 0.01$).

Ileal and fecal concentration of crude mucin increased from the LSF to the HSF diet by 78 and 46% ($P < 0.01$; Table 3). In addition, the ileal crude mucin flow (g DM/d) of the rabbits fed the HSF was higher than that of rabbits fed the LSF diet ($P = 0.004$). A similar effect, but smaller in magnitude, was observed for the fecal crude mucin flow when it was expressed per kilogram of DMI ($P = 0.031$). The apparent cecal crude mucin fermentation of the rabbits fed the HSF diet was higher than that of rabbits fed the MSF and LSF diets (90.9 vs. 84.8%; $P = 0.014$). Ileal and fecal crude mucin contained 24.3 and 21.6% CP.

Rabbits fed the LSF diet showed an ileal CP digestibility 8% higher than rabbits fed the other 2 diets (64.9 vs. 59.8%; $P < 0.01$; Table 4). These values increased by 3.7 percentage units when they were corrected for the ileal mucin CP (59.7 vs. 63.4; $P = 0.002$). No difference was found in cecal CP digestibility among rabbits fed the three experimental diets, and it decreased by 3.3% units when the mucin CP was considered (12.8 vs. 9.52%; $P = 0.006$). Fecal CP digestibility of rabbits fed the LSF diet was 8% higher than that of rabbits fed the MSF diet ($P = 0.002$), with the HSF diet generating an intermediate value. Once

Table 2. Effect of type of fiber and site of digestion on apparent ileal, cecal, and fecal digestibility of dietary components in rabbits

Item	DM	OM	Starch	GE
Ileal digestibility, %				
Low soluble fiber	48.9	50.9	98.3	–
Medium soluble fiber	46.2	48.2	98.1	–
High soluble fiber	52.1	54.6	98.0	–
Cecal digestibility, %				
Low soluble fiber	16.2	13.9	1.15	–
Medium soluble fiber	18.2	15.7	1.52	–
High soluble fiber	18.4	17.8	1.33	–
SEM[1]				–
Type of fiber (diet)	0.42	0.54	0.064	–
Site of digestion	1.62	1.81	0.217	–
Type of fiber × site of digestion	1.99	2.25	0.269	–
P-value				
Type of fiber (diet)	<0.001	<0.001	0.31	–
Site of digestion	<0.001	<0.001	<0.001	–
Type of fiber × site of digestion	0.574	0.726	0.693	–
Fecal digestibility, %				
Low soluble fiber	65.1[b]	64.8[b]	99.5	65.6[b]
Medium soluble fiber	64.4[b]	63.9[b]	99.6	64.3[b]
High soluble fiber	70.5[a]	72.4[a]	99.3	69.7[a]
SEM[1]	0.83	1.08	0.13	0.83
P-value				
Type of fiber (diet)	<0.001	<0.001	0.267	<0.001

[a,b]Mean values in the same column for each site of digestion with a different superscript differ ($P < 0.05$).

[1]n = 10 rabbits/diet in each site of digestion.

corrected for fecal mucin CP, fecal CP digestibility increased by 0.4% units ($P < 0.01$).

Ileal digestibility of TDF increased from the LSF to the HSF diet group (10.5 vs. 25.6%; $P < 0.01$; Table 4), but the cecal digestibility of TDF was unaffected by the diet (28.2% on average), resulting in an interaction diet × site of fermentation ($P < 0.01$). Ileal TDF digestibility increased by 4.2% units when it was corrected for mucins ($P < 0.01$), whereas cecal digestibility decreased by 3.7% units ($P < 0.01$). As a result, an interaction mucin correction × site of fermentation was found ($P < 0.01$). Once corrected for mucin content, around 40% of digestible TDF was fermented in the ileum and 60% in the cecum (18.5 vs. 26.4%, respectively; $P < 0.01$). However, it depended on the type of diet because the increment of dietary soluble fiber content raised the proportion of mucin corrected TDF fermented in the ileum (from 32 to 51% for the LSF to the HSF diets, respectively; $P < 0.01$). Accordingly, rabbits fed the HSF diet fermented daily in the ileum more than double the TDF of those fed the MSF and LSF diets (16.7 vs. 7.7 g TDF/d; $P < 0.01$; Fig. 1) but a similar amount in the cecum (16.2 g TDF/d). Rabbits fed the MSF and LSF diets fermented double amount of TDF in the cecum than in the ileum (15.3 vs. 7.7 g TDF/d; $P < 0.01$; Fig. 1). Fecal digestibility of TDF raised from 37.5 to 54.7% for the LSF and HSF diet groups, respectively ($P < 0.01$). Moreover, it increased by 0.5% units once corrected for fecal mucin ($P < 0.01$).

Ileal digestibility of insoluble fiber of rabbits fed the HSF was greater than for those fed the LSF diet (21.0 vs. 11.3%; $P < 0.01$; Table 5), but cecal digestibility was similar among the 3 diet groups (13.9% on average), leading to an interaction between type of fiber × site of fermentation ($P = 0.013$). No effect of the methods used to determine insoluble fiber was found on its digestibility

Table 3. Effect of type of fiber (low soluble fiber, medium soluble fiber, or high soluble fiber diet) on ileal and fecal mucin content in rabbits

	Type of fiber				
Item	Low soluble	Medium soluble	High soluble	SEM[1]	*P*-value
Ileal crude mucin					
g DM/kg dry ileal digesta	43.3[c]	59.7[b]	77.1[a]	4.41	<0.001
g DM/d	3.20[b]	3.80[b]	5.64[a]	0.48	0.004
g/kg of DMI	21.4[c]	28.2[b]	42.5[a]	3.00	<0.001
CP, % ileal mucin	25.3	24.3	23.2	0.70	0.136
Fecal crude mucin					
g DM/kg of dry fecal digesta	8.44[b]	11.0[a]	12.3[a]	0.62	<0.001
g DM/d	0.45	0.55	0.50	0.062	0.502
g/kg of DMI	2.94[b]	3.95[a]	3.66[ab]	0.26	0.031
CP, % fecal mucin[2]	22.6	21.4	20.8	–	–
Apparent cecal fermentation of mucin, %	85.2[b]	84.5[b]	90.9[a]	1.53	0.014

[a–c]Mean values in the same row with a different superscript differ ($P < 0.05$).

[1]n = 10 rabbits/diet.

[2]CP on fecal mucin was determined using only 1 pooled sample for each treatment due to the small residue obtained.

Table 4. Effect of type of fiber (low soluble fiber [LSF], medium soluble fiber [MSF], or high soluble fiber [HSF] diet), mucin correction, and site of digestion/fermentation on apparent ileal, cecal, and fecal digestibility of CP and total dietary fiber in rabbits

	CP		Total dietary fiber	
Mucin correction	No	Yes	No	Yes
Ileal digestibility, %				
LSF	63.5[a]	66.3[a]	8.91[b]	12.0[b]
MSF	57.6[b]	61.5[b]	10.9[b]	15.5[b]
HSF	57.9[b]	62.3[b]	23.0[a]	28.1[a]
Average	59.7	63.4	14.3	18.5
Cecal digestibility, %				
LSF	11.9	9.43	28.4	25.8
MSF	12.0	8.53	30.5	26.6
HSF	14.5	10.6	31.4	26.9
Average	12.8	9.52	30.1	26.4
SEM[1]				
Type of fiber (diet)	0.57		0.68	
Mucin correction/site of digestion[2]	0.53		0.56	
Fiber method	–		–	
Type of fiber × site of digestion	0.91		0.96	
Mucin correction × site of digestion	0.79		0.79	
P-value[3]				
Type of fiber (diet)	0.008		<0.001	
Mucin correction	0.817		0.742	
Site of digestion	<0.001		<0.001	
Type of fiber × site of digestion	0.004		<0.001	
Mucin correction × site of digestion	<0.001		<0.001	
Fecal digestibility, %				
LSF	75.4[a]	75.7[a]	37.3[c]	37.7[b]
MSF	69.6[b]	70.0[b]	41.4[b]	42.0[b]
HSF	72.5[ab]	72.9[ab]	54.5[a]	55.0[a]
Average	72.5	72.9	44.4	44.9
SEM[1]				
Type of fiber (diet)	1.15		1.36	
Mucin correction	0.670		0.803	
Type of fiber × mucin correction	1.15		1.36	
P-value[3]				
Type of fiber (diet)	0.008		<0.001	
Mucin correction	<0.001		<0.001	
Type of fiber × mucin correction	0.097		0.064	

[a,b]Mean values in the same column and site of digestion/fermentation for each effect with a different superscript differ ($P < 0.05$).

[1]n = 10 rabbits/diet. Standard error of the mean values for interaction type of fiber × mucin correction × site of digestion/fermentation for ileal and cecal CP and total dietary fiber digestibility were 1.35 and 1.34, respectively.

[2]Standard error of the mean values for mucin correction and site of digestion/fermentation were the same for CP and total dietary fiber.

[3]The interactions not shown were not significant ($P > 0.20$).

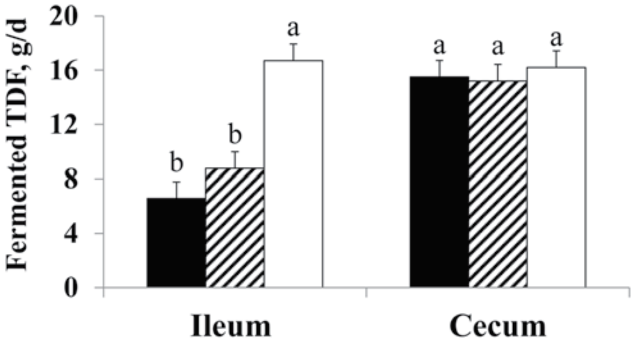

Figure 1. Effect of type of fiber (low [■], medium [▨], or high soluble fiber diet [□]) and site of fermentation (ileum vs. cecum) on the daily total dietary fiber (TDF) fermented (g TDF/d, corrected for mucin content and considering the DMI as covariate). P_{type} of fiber × site of fermentation < 0.001. Values are means ± SEM (0.79; n = 10). [a,b]Within each segment mean values with a different letter differ ($P < 0.05$).

at the ileum or cecum. Ileal and cecal digestibility of insoluble fiber showed on average similar values (15.7 vs. 13.9%; $P = 0.078$). According to these results, approximately half of the total fermented insoluble fiber (53%) occurred before the cecum. Consequently, the fecal digestibility of insoluble fiber was greater for rabbits fed the HSF diet than for those fed the MSF and LSF diets (37.7 vs. 25.5%; $P < 0.01$; Table 4). Fecal digestibility of insoluble fiber measured as NDF was higher than that measured as IDF or in vitro insoluble fiber (31.4 vs. 28.7%; $P < 0.01$). The amounts of NDF, IDF, and in vitro insoluble fiber fermented (g/d) in the whole digestive tract were highly correlated ($r \geq 0.973$, $P < 0.01$). This correlation remained high between the amount of NDF and IDF fermented, in both the ileum and cecum ($r \geq 0.829$, $P < 0.01$). In contrast, the correlation between the in vitro insoluble fiber and the NDF or IDF fermented in the ileum and cecum decreased (from 0.445 to 0.569; $P < 0.05$) or even disappeared (between in vitro insoluble fiber and IDF fermented in the cecum).

The rabbits fed the HSF diet had a greater ileal digestibility of soluble fiber than those fed the MSF and LSF diets (35.6 vs. 7.6%, respectively; $P < 0.01$; Table 5). It was 16.5% units greater when it was corrected using the ileal mucin content than the uncorrected value (8.69 vs. 25.2%; $P < 0.01$). When the ileal soluble fiber digestibility was not corrected for mucin, it showed negative values for rabbits from the LSF and MSF diet groups (–0.40 and –1.13%), being positive only for those of the HSF diet group (27.6%). Ileal digestibility of soluble fiber became positive for rabbits fed the LSF and MSF diets after its correction for mucin content (14.5 and 17.3%), whereas it showed a higher value for those of HSF diet group (43.6%). Once corrected for mucin content, the ileal digestibility of soluble fiber for each diet group was not affected by the fiber methodology used. Ileal digestibil-

Table 5. Effect of type of fiber (low soluble fiber [LSF], medium soluble fiber [MSF], or high soluble fiber [HSF] diet), mucin correction, fiber method, and site of fermentation on apparent ileal, cecal, and fecal digestibility of insoluble and soluble fiber in rabbits

	Insoluble fiber			Soluble fiber					
Fiber method[1]	Insoluble dietary fiber	In vitro insoluble fiber	NDF	Total dietary fiber–insoluble dietary fiber		Total dietary fiber–in vitro insoluble fiber		Total dietary fiber–NDF	
Mucin correction				No	Yes	No	Yes	No	Yes
Ileal digestibility, %									
LSF	12.1[b]	10.7[b]	11.0[b]	–5.04[b]	11.4[b]	2.86[b]	16.2[b]	0.98[b]	15.9[b]
MSF	15.7[ab]	14.8[ab]	14.1[b]	–3.78[b]	14.9[b]	–1.02[b]	17.5[b]	1.42[b]	19.6[b]
HSF	20.7[a]	20.9[a]	21.4[a]	28.5[a]	45.2[a]	27.9[a]	44.4[a]	26.4[a]	41.4[a]
Average	16.2	15.5	15.5	6.55	23.8	9.92	26.0	9.59	25.7
Cecal digestibility, %									
LSF	12.6	13.2	15.4	97.4[a]	83.2[a]	79.0[a]	67.5[a]	78.6[a]	65.6[a]
MSF	9.56	10.1	14.1	95.7[a]	79.4[a]	93.5[a]	77.4[a]	79.9[a]	64.1[a]
HSF	16.2	15.9	18.2	66.6[b]	51.6[b]	66.6[b]	51.8[b]	57.7[b]	44.2[b]
Average	12.8	13.0	15.9	86.5	71.4	79.7	65.6	72.1	58.0
SEM[2]									
Type of fiber (diet)		0.92				0.51			
Mucin correction/site of fermentation[3]		0.72				1.05			
Fiber method		0.88				1.45			
Type of fiber × site of fermentation		1.26				1.78			
Mucin correction × site of fermentation		–				1.76			
Fiber method × site of fermentation		1.26				2.62			
P-value[4]									
Type of fiber (diet)		<0.001				<0.001			
Mucin correction		–				0.61			
Fiber method		0.462				0.057			
Site of digestion		0.078				<0.001			
Type of fiber × site of fermentation		0.013				<0.001			
Mucin correction × site of digestion		–				<0.001			
Fiber method × site of fermentation		0.284				0.004			
Fecal digestibility, %									
LSF	24.7[b]	23.9[b]	26.4[b]	92.4	94.6	81.9[b]	83.7[b]	79.5[b]	81.6[b]
MSF	25.2[b]	24.9[b]	28.2[b]	91.9	94.3	92.5[a]	94.9[a]	81.4[ab]	83.7[ab]
HSF	36.9[a]	36.8[a]	39.6[a]	95.1	96.7	94.6[a]	96.2[a]	84.1[a]	85.6[a]
Average	28.9[B]	28.5[B]	31.4[A]	93.1	95.2	89.7	91.6	81.7	83.6
SEM[2]									
Type of fiber (diet)		1.84				1.02			
Mucin correction		–				0.64			
Fiber method		1.09				0.67			
Type of fiber × mucin correction		–				1.09			
Type of fiber × fiber method		1.86				1.15			
P-value[4]									
Type of fiber (diet)		<0.001				<0.001			
Mucin correction		–				<0.001			
Fiber method		<0.001				<0.001			
Type of fiber × fiber method		0.503				<0.001			

[a,b]Mean values in the same column and site of fermentation for each effect with a different superscript differ ($P < 0.05$).

[A,B]Average values in the same row for insoluble fiber with a different superscript differ ($P < 0.05$).

[1]$n = 10$ rabbits/diet. Standard error of the mean values for interaction type of fiber × fiber method × site of fermentation for ileal and cecal insoluble and soluble fiber digestibility were 2.13 and 3.85, respectively.

[2]Standard error of the mean value for interaction type of fiber × mucin correction × fiber method × site of fermentation for ileal and cecal insoluble fiber digestibility was 5.68. Standard error of the mean value for interaction type of fiber × mucin correction × fiber method for fecal soluble fiber digestibility was 1.31.

[3]Standard error of the mean values for mucin correction and site of fermentation were the same for soluble fiber digestibility.

[4]The interactions not shown were not significant ($P > 0.20$).

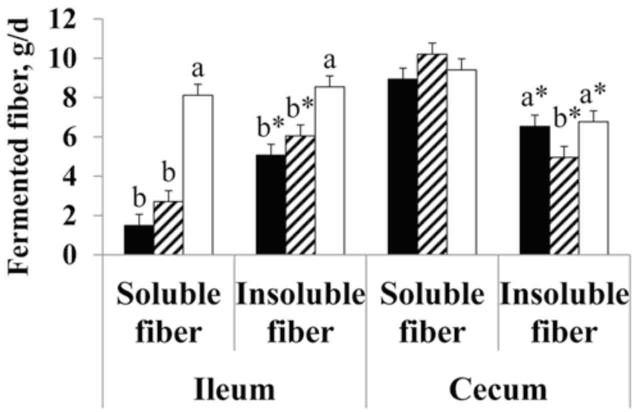

Figure 2. Effect of type of fiber (low [■], medium [▨], or high soluble fiber diet [□]), site of fermentation (ileum vs. cecum), and fiber fraction (soluble vs. insoluble) on daily fiber fermented (g/d, corrected for mucin content and considering the DMI as covariate). Values for fermented soluble and insoluble fiber are the average of the 3 methods evaluated. $P_{\text{type of fiber × site of fermentation × fiber fraction}}$ = 0.002. Values are means ± SEM (0.51; n = 10). [a,b]Within each combination type of fiber × site of fermentation mean values with a different letter differ (P < 0.05). *Within each segment indicates that mean fiber fraction values for each type of fiber differ (P < 0.05).

Table 6. Effect of type of fiber (low soluble fiber, medium soluble fiber, or high soluble fiber diet) on ileal flow (g/d) of dietary components in rabbits

	Type of fiber				
Item	Low soluble	Medium soluble	High soluble	SEM[1]	P-value
DM	71.9	77.5	69.6	2.57	0.108
OM	64.6	69.0	61.4	2.46	0.119
CP	10.0[b]	11.7[a]	11.8[a]	0.40	0.006
CP-mucin[2]	0.75[b]	1.07[a]	1.22[a]	0.074	<0.001
CP-NDF[3]	6.47	5.42	4.80	0.57	0.150
Starch	0.51	0.58	0.60	0.078	0.706
TDF,[4] mucin corrected	51.7[a]	49.2[b]	45.1[c]	0.62	<0.001

[a–c]CP, CP-mucin, CP-NDF, Starch and TDF-mucin corrected should be moved to the left. Mean values in the same row with a different superscript differ (P < 0.05).

[1]n = 10 rabbits/diet. Covariate DMI was significant (P < 0.01) for all traits.

[2]CP from intestinal mucin. DF = total dietary fiber.

[3]CP linked to NDF.

[4]Total dietary fiber corrected for mucins.

ity of soluble fiber (corrected for mucin) was higher than that for insoluble fiber for rabbits fed the HSF diet (43.6 vs. 21.0%; P < 0.01), and no difference was detected for animals who received the LSF and MSF diets (15.9 vs. 13.1%). As a consequence, the amount of fermented soluble fiber (once corrected for mucin content) in the ileum was lower than the fermented insoluble fiber for rabbits fed the LSF diet (1.5 vs. 5.1 g/d; P < 0.01; Fig. 2) and the MSF diet (2.7 vs. 6.0 g/d; P < 0.01), but it was similar for those given the HSF diet (8.1 vs. 8.5 g/d). In contrast to the ileal digestibility, cecal digestibility of soluble fiber was lower for the HSF compared with the MSF and LSF diet groups (56.4 vs. 80.1%; P < 0.01), resulting in an interaction diet × site of fermentation (P < 0.01). Correction for ileal and fecal mucin content decreased cecal soluble fiber digestibility by 14.4% units compared with the uncorrected value (P < 0.01). This effect was opposite to that recorded in the ileum and led to an interaction mucin correction × site of fermentation (P < 0.01). Once corrected for ileal and fecal mucin content, digestibility of soluble fiber in the cecum was higher than in the ileum (65.0 vs. 25.2%; P < 0.01). However, it depended on the type of diet because the increment of dietary soluble fiber content raised the proportion of soluble fiber fermented in the ileum (from 14.5 to 43.6% for the LSF and HSF diet groups, respectively; P < 0.01) but decreased it in the cecum (72.1 vs. 49.2% for the HSF and the average of the MSF and LSF diet groups, respectively; P < 0.05). As a consequence, the amount of soluble fiber fermented in the cecum was greater than in the ileum for rabbits who received the LSH and MSF diets (9.5 vs. 2.1 g/d; P < 0.01; Fig. 2) but similar for those given the HSF diet (9.4 vs. 8.1 g/d). The method used to quantify soluble fiber did not affect its ileal digestibility but influenced its cecal digestibility, being lower for TDF–NDF than for the other 2 methods (65.0 vs. 75.8%; P = 0.031), and consequently the interaction fiber method × site of fermentation was significant (P = 0.004). The fecal digestibility of soluble fiber increased with the dietary soluble fiber level (P < 0.001; Table 5). Moreover, its value decreased successively (P < 0.01) when it was determined as TDF–IDF, TDF–in vitro insoluble fiber, and TDF–NDF. An interaction diet × method was found for fecal digestibility of soluble fiber (P < 0.01; Table 5), because it was lower for the LSF compared with the HSF diet group, but for TDF–IDF, rabbits fed LSF and HSF diets showed similar values. Once corrected for fecal mucin content, fecal digestibility of soluble fiber increased by 2.0% units (P < 0.01). The amounts of fermented TDF–NDF, TDF–IDF, and TDF–in vitro insoluble fiber in the whole digestive tract were highly correlated ($r \geq 0.969$, P < 0.01). This correlation remained high between the amount of fermented TDF–NDF and TDF–IDF, in both the ileum and cecum ($r \geq 0.830$, P < 0.01). However, the correlation among the amount of fermented TDF-in vitro insoluble fiber and TDF-NDF or TDF-IDF in the ileum decreased (from 0.541 to 0.580; P < 0.05) or even disappeared in the cecum. All the interactions not mentioned were not significant.

Rabbits fed the LSF diet showed the lowest ileal flow of total and mucin CP (P < 0.01; Table 6). No effect was observed in the ileal flow of DM, OM, CP linked to NDF, and starch, but the TDF ileal flow (cor-

Table 7. Effect of type of fiber (low soluble fiber [LSF], medium soluble fiber [MSF], or high soluble fiber [HSF] diet) and fiber method on ileal flow (g/d) of dietary insoluble fiber and soluble fiber, corrected for ileal mucin content, in rabbits

Item	Insoluble fiber	Soluble fiber
Diets		
LSF	41.3[a]	11.9
MSF	37.6[b]	10.8
HSF	34.0[c]	11.5
Method[1]		
IDF	38.0	11.0
In vitro insoluble fiber	37.5	11.2
NDF	37.4	11.2
SEM[2]	0.92	0.81
P-value		
Type of fiber (diet)	<0.001	0.248
Fiber method	0.711	0.637
Type of fiber × fiber method	0.624	0.513

[a–c]Mean values in the same column for each site of fermentation with a different superscript differ ($P < 0.05$).

[1]IDF = insoluble dietary fiber; NDF = **α**-amylase NDF corrected for ash and protein.

[2]$n = 10$. Covariate DMI was significant ($P < 0.01$) for all traits.

rected for mucin) decreased when the soluble fiber increased ($P < 0.01$). Independent of the method used, the ileal flow of insoluble fiber was higher for rabbits fed the LSF compared with the HSF diet group (41.3 vs. 34.0 g/d; $P < 0.01$; Table 7). Ileal flow of soluble fiber (corrected for ileal mucin) was unaffected by the diet and by the method of analysis (11.1 g/d, on average).

DISCUSSION

This work confirms that increasing soluble fiber (by the inclusion of sugar beet and apple pulp) improves fecal TDF digestibility as reported previously (Trocino et al., 2011; Xiccato et al., 2011). The dietary soluble fiber represents around 50% of the total fermented TDF, despite its low proportion in the diet, and it is independent of the diet. This is explained by both the high fecal digestibility of soluble fiber, which is very close to that of starch (91.4 vs. 99.5%, respectively), and the improved fecal digestibility of insoluble fiber when the soluble fiber increases. Ingredients rich in soluble fiber (sugar beet or citrus pulps) showed greater fecal digestibility of insoluble fiber than other common sources of fiber such as wheat bran, dehydrated alfalfa, or straw, which is mainly caused by their low degree of lignification (Martínez Pascual and Fernández Carmona, 1980; Gidenne, 1987; Pérez de Ayala et al., 1991).

The identification of the site of fermentation of soluble fiber and TDF (or nonstarch polysaccharides) is complex because of their usually low or even negative ileal digestibility found in this study or in previous work performed in pigs (Graham et al., 1986; Jørgensen et al., 1996) and rabbits (Gidenne, 1992; Martínez-Vallespín et al., 2013). This circumstance has been related in pigs with a potential interference of endogenous substances with these determinations (Graham et al., 1986; Jørgensen et al., 1996; Wilfart et al., 2007). In fact, Abad et al. (2013) observed that rabbit intestinal mucins were retained within the soluble fiber fraction when the latter was quantified in the intestinal digesta as both precipitate with ethanol. Consequently, a correction to avoid this problem was proposed. After applying this correction in the current study, the ileal TDF and soluble fiber digestibility increased. In addition, soluble fiber digestibility became positive for rabbits fed the diets with the lowest soluble fiber content.

Ileal digestibility of TDF (corrected for mucins) represented on average 40% of the fecal TDF digestibility and it was positively influenced by soluble fiber inclusion. This value is similar to that obtained previously in pigs (Mathers, 1991) or rabbits (Gidenne, 1992; García et al., 1999; Carabaño et al., 2001), when the digestibilities of dietary fiber monomers were determined. The increase of soluble fiber in the diet improved the ileal digestibility of insoluble and soluble fiber (corrected for mucins). The amount of fermented insoluble fiber in the ileum was higher than the amount of fermented soluble fiber (once corrected for mucins) for rabbits who received the LSF and MSF diets (72 vs. 28% out of the total ileal fermented TDF). When the soluble fiber content of the diet increased, as in the HSF diet, the rate of fermentation in the ileum was similar for insoluble and soluble fiber (52 vs. 48% out of the total ileal fermented TDF). These results are explained by the higher proportion of insoluble fiber in the diet (especially in the LSF and MSF diets) and the similar ileal digestibility of insoluble and soluble fiber. However, the amount of insoluble fiber fermented in the ileum is surprisingly high taking into account the relative short oroileal mean retention time of the digesta (around 5 h; Gidenne, 1994; García et al., 1999). In addition, a higher ileal digestibility of the soluble fiber fraction compared to the insoluble fiber fraction would have been expected because it is easily available and the pectinase activity in the rabbit small intestine is higher than the xylanase and cellulase activity (Marounek et al., 1995). A minor proportion of the insoluble fiber digestibility in the ileum may be explained by its partial solubilization in the acid conditions of the stomach, as observed when determining the 2-step pepsin/pancreatin in vitro NDF digestibility (from 6 to 1% for the HSF diet and the average of the LSF and HSF diets, respectively; unpublished data). Similarly, a high ileal NDF digestibility was also found in rabbits given diets containing dehydrated alfalfa (50%) or dehydrated alfalfa and sugar beet pulp (20 and 30%) as the main sources of fiber (39 and 43%, respectively;

Merino and Carabaño, 1992). Other authors also reported a positive insoluble fiber digestibility before the cecum in rabbits (from 7 to 19% for crude fiber [Yu et al., 1987], 5% for NDF [Gidenne and Ruckebusch, 1989], and 16% for NDF [Blas et al., 2003]), ponies (13% for NDF; Hintz et al., 1971), and pigs (from negative values to 43% for NDF [Keys and DeBarthe, 1974], from 7 to 53% for IDF [Graham et al., 1986], and 17% for NDF [Schulze et al., 1994]). These results seem to confirm the occurrence of a significant hydrolysis of cell wall polysaccharides (soluble and insoluble) in the ileum, which might be performed by extracellular fibrolytic enzymes (Abbott and Boraston, 2008), as observed in the rabbit cecum (Sirotek et al., 2001). It might suggest that the cell wall polysaccharides in the ileum can be hydrolyzed to carbohydrates with lower molecular weight (to shorter polysaccharides or to oligosaccharides) as opposed to being extensively fermented. In fact, the molecular weight of β-glucan of oats was reduced after passage through the stomach and the proximal small intestine of pigs. It was probably associated to the microbial enzymes, although this activity did not lead to a loss in β-glucan from the digesta before the lower small intestine was reached (Johansen et al., 1993). Consequently, low molecular weight carbohydrates derived from microbial fiber hydrolisis would not be retained in the insoluble fiber fraction or precipitated with ethanol. This result may be explained by the fact that some bacteria possesses polysaccharide depolymerases but not glycoside hydrolases necessary for utilization of the xylooligosaccharides or oligogalacturonides as an energy source leading to the solubilization but not fermentation of cell wall polysaccharides (Dehority, 1993). In this way, Fondevila and Dehority (1994) demonstrated that ruminal hemicellulose utilization resulted from initial solubilization of the hemicellulose from the forage by nonhemicellulose utilizers and subsequent fermentation of this soluble polysaccharide by the utilizing, but nondegrading, bacteria. This circumstance may occur in rabbits fed high soluble fiber diets and may affect the ileal microbiota profile and gut health. El Abed et al. (2013) reported a reduction in ileal biodiversity in rabbits fed a diet including sugar beet pulp, although this effect was not observed by Gómez-Conde et al. (2007).

The majority of TDF was fermented in the cecum (60%, which corresponds to 15.9 g TDF/d, on average), and the TDF fermented in the cecum was not affected by dietary treatments. The soluble fiber represented 61% of the total TDF fermented in the cecum (9.7 g/d, on average). This was in agreement with the higher fibrolytic activity found in the cecum, which was around 8.5 times higher than in the ileum (Marounek et al., 1995), and the longer mean retention time in the cecum and colon (around 9 h; Gidenne et al., 2010). The most important cecal fibrolytic activity found in rabbits fed alfalfa- and sugar beet pulp–based diets was the pectinase activity, whereas the cellulolytic activity was the least important (Falcao e Cunha et al., 2004). These authors observed that cecal pectinolytic activity was overcome only by xylanolytic activity in rabbits fed a wheat bran–based diet.

The method for quantifying insoluble fiber had no influence on the ileal and cecal digestibility of this fraction or on the amount of fermented insoluble fiber in each segment. However, its fecal digestibility was slightly higher when using NDF compared with IDF or in vitro insoluble fiber, leading to a higher amount of insoluble fiber fermented when determined using NDF respect to the value obtained with in vitro insoluble fiber (13.3 vs. 12.1 g insoluble fiber/d; $P = 0.023$). These differences are not relevant and might be explained by the higher solubilization of hemicelluloses in the feces than in the ingredients when the detergent method is used, compared with an enzymatic-chemical method, as reported by Hindrichsen et al. (2006). In fact, the correlation among the amount of insoluble fiber fermented in the whole digestive tract using the 3 methods was high. This correlation remained high for the insoluble fiber fermented in the ileum and cecum when it was calculated as NDF and IDF, but the TDF fermented in the cecum decreased for the insoluble fiber fermented in the cecum when in vitro insoluble fiber was used or even disappeared in the ileum. Similarly, the method for quantifying soluble fiber had no influence on the quantification of its ileal digestibility but influenced the cecal and fecal digestibility of this fraction, being lower for TDF–NDF digestibility. Nevertheless, for practical purposes, the method used to determine soluble fiber digestibility had no effect on the amount of soluble fiber fermented either in the ileum or in the cecum. The differences obtained for fecal soluble digestibility among diet groups due to the method used did not change the differences between the extreme diets, but it would suggest that the differences in the digestibility of extreme diets might increase when the soluble fiber digestibility is estimated using TDF–in vitro insoluble fiber. The correlation among the amount of soluble fiber fermented in the whole digestive tract using the 3 methods was high. This correlation remained high in the ileum and cecum when the soluble fiber fermented was calculated as NDF and IDF, but this correlation decreased for the soluble fiber fermented in the ileum and cecum when in vitro insoluble fiber was used. This result may be related to the different recovery of solubilized (partial or total) polysaccharides by the insoluble fiber methods. Accordingly, the nutritional meaning of the 3 different methodologies does not differ at the fecal level (especially when the amount of fermented fiber is considered). However, the meaning of the values obtained with the in vitro insoluble fiber method might differ from those obtained with NDF and IDF at both ileal and cecal levels.

The observed positive effect of soluble fiber on ileal mucin flow agrees with the higher number of goblet cells per villi found in the jejunum when the soluble fiber is increased in rabbits (El Abed et al., 2011) and rats (Ito et al., 2009). The minor differences detected in the mucin flow at the fecal level indicated that mucins are extensively fermented by cecal bacteria, as observed in vitro by Marounek et al. (2000). In fact, the amount of mucins fermented in the cecum of rabbits fed the HSF diet represented around 50% of the amount of soluble fiber fermented (5.0 vs. 9.8 g/d), whereas this proportion decreased for those fed the LSF diet (2.7 vs. 9.0 g/d). The potential effects exerted by the inclusion of soluble fiber through raising the mucin production in the intestinal mucosa or in the cecum, as a substrate for microbial fermentation, might be related to the positive effect of soluble fiber on mortality rate in growing rabbits (Martínez-Vallespín et al., 2011; Trocino et al., 2013).

In conclusion, the ileal and cecal digestibility of TDF and soluble fiber should be corrected for the ileal and fecal mucin content. The method selected to measure insoluble and soluble fiber did not have a relevant influence on the amount of the fermented fiber fractions. Additionally, the increment of soluble fiber using sugar beet and apple pulp increased the amount of TDF fermented, due to the increasing proportions of both the insoluble and soluble fiber fractions fermented/solubilized in the ileum.

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